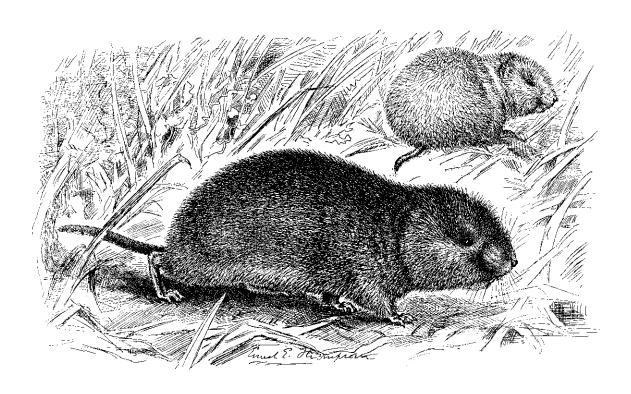
Mammal Inventory of Alaska's National Parks and Preserves

Yukon-Charley Rivers National Preserve

Annual Report 2001



University of Alaska Museum

Stephen O. MacDonald, Research Associate

Joseph A. Cook, Principal Investigator

28 December 2001

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Executive Summary

This project was a cooperative effort of the University of Alaska Museum (UAM), the Inventory and Monitoring Program of the National Park Service (NPS) of Alaska, and the Beringian Coevolution Project at Idaho State University. Participating in this project were scientists from the U.S. National Parasite Lab-Beltsville, Maryland, the University of New Mexico, the Harvard School of Public Health, Institute of Biological Problems of the North-Magadan, Russia, Vantaa Research Centre-Vantaa, Finland, and the University of Saskatchewan-Saskatoon, Canada.

This report details the inventory of the mammals at 10 general localities in the Yukon-Charley Rivers National Preserve (YUCH, the Preserve), upper Yukon River region, Alaska, in July and August 2001. We begin the process of documenting the approximately 40 species of mammals that occur in YUCH, with a primary focus on small mammals (the shrews, voles, lemmings, mice, weasels, porcupine, squirrels, pika and hare).

This survey (328 person days and approximately 13,000 trap nights of sampling effort) resulted in 1891 primary specimens which documented 17 small mammal species. A review of specimen holdings in other major collections bring the total number of documented small mammal species in YUCH to 21 (out of a probable 24).

This inventory included a rigorous protocol for the physical documentation of the data including the systematic collection of diverse preparations of museum specimens. In addition to standard mammal preparations (fluid preserved, skin, and skeletal preparations), we archived ultrafrozen tissues (heart, liver, kidney) for future toxicology, genetic or stable isotope research. We also preserved a significant series of ectoparasites (e.g., fleas, ticks, lice), endoparasites (e.g., helminth worms), blood borne parasite preparations (e.g., babesia), and other protozoan preparations (e.g., coccidian) or viral (e.g., hantavirus) preparations. Much of this material is currently under investigation by a number of laboratories worldwide and will result in a more holistic view of the mammalian diversity in YUCH. Perspectives on the value of the specimenbased approach to inventory and monitoring are discussed, and recommendations for future efforts are enumerated.

A few immediate highlights from the field work include 10 specimens of the tiny shrew (Sorex yukonicus). These records nearly doubles the number known to science and extend the known distribution of this newly described species significantly. Other specimens from this survey have already been incorporated into ongoing molecular genetic studies. For example, Dr. Vadim Fedorov has identified the presence of two distinct phylogeographic lineages of brown lemmings (Lemmus trimucronatus) within YUCH. These results, combined with other recent work on ermine, suggests that the Preserve may be an important zone of contact for a number of species and hence harbor substantially more diversity than currently suspected. These preliminary discoveries based on the field work and specimens suggest that significant new information will be forthcoming regarding our understanding of the mammalian fauna of YUCH.

Introduction

This report details an inventory of the small mammals of the Yukon-Charley Rivers National Preserve in July and August 2001. The University of Alaska Museum and the Beringian Coevolution Project at Idaho State University (ISU) worked collaboratively with the Inventory and Monitoring Program of the National Park Service to conduct an inventory at selected sites throughout the Yukon-Charley Rivers National Preserve to document the occurrence, relative abundance, and general habitat affinities of the small mammal fauna. This effort provided a large series and variety of permanently preserved materials and associated data sets for taxonomic, zoogeographic, ecological, genetic, parasitological, epidemiological, and other research and management purposes.

The documentation of species' occurrence in the Preserve was complemented by a review of specimen holdings at the University of Alaska Museum (UAM) and other major collections, primarily the U.S. National Museum (USNM).

Scientific and common names of mammals used in this report follow Wilson and Reeder (1993) and Wilson and Cole (2000), respectively. Vegetation classification generally follows Viereck et al. (1992)

Methods and Materials

Two separate field crews sampled 10 base locations (Figure 1) for a total of 328 person-days (approximately 13,000 trapnights of collecting effort). The sites were scattered throughout the YUCH in a variety of tundra- and taiga-zone habitats at various elevations. Logistical support was provided by the National Park Service (Fairbanks and Eagle).

Field Locations

Upper Crescent Creek (64°48.7'N, 143°46.0'W). 15 – 21 July 2001. Camp was established at 1321 m in the mountains of the Upper Charley River Watershed next to a small, unnamed alpine lake at the headwaters of Crescent Creek.

Copper Mountain (64°36.6'N, 143°06.0'W). 21 - 24 July 2001. Camp was located at 1006 m in an expansive area of open tundra in the far upper reaches of Charley River, approximately 9 km SE of the "Copper Mountain" benchmark.

Mount Sorenson (64°58.1'N, 143°03.0'W). 24 – 29 July 2001. This alpine field camp was located at 1037 m, approximately 4.5 km SW of Mt. Sorenson, at the uppermost headwaters of Seventymile River.

Mount Kathryn (65°10'N, 143°33.0'W). 30 July – 4 August 2001. Located about 3.6 km SW of Mt. Kathryn, this field camp was situated at 1220 m on the alpine ridge separating the upper watersheds of Woodchopper Creek and Coal Creek.

Squaw Mountain $(65^{\circ}03.7^{\circ}N, 141^{\circ}00^{\circ}W)$. 16-21 July 2001. This steep alpine field camp was established at 1131 m on a narrow ridge just above treeline near the International Border. Traplines were established in a variety of subalpine forest to alpine tundra habitats that ranged in elevation from 977 m to 1183 m.

Kathul Mountain ($65^{\circ}22.3$ 'N, $142^{\circ}01.1$ 'W). 21 - 30 July 2001. Field camp was located about 5 km N of the Yukon River and 11 km ENE of Kathul Mt., in a low saddle just above

treeline (955 m elevation). A variety of forest to alpine habitats was sampled at elevations ranging from 850 m to 1050 m.

Upper Kandik River below Johnson Gorge (65°26.4'N, 142°00.5'W). 30 July – 4 August 2001. Our tent camp was situated at the edge of an open boggy area in forest (363 m elevation) about 1 km S of the Kandik River just below Johnson Gorge. A variety of river bottom and upland habitats were sampled. The mosaic of wetlands habitats of an active beaver colony at the edge of the Kandik's floodplain received particular attention.

Yukon River, Slaven Cabin area $(65^{\circ}21.0^{\circ}N, 143^{\circ}07.3^{\circ}W)$. 4-18 August 2001. Slaven Cabin was used as a base of operations to survey by boat and 4-wheeler areas along the Yukon River and Coal Creek. A wide variety of lowland forest, scrub and herbaceous habitats were sampled within a 12 km radius of camp.

Yukon River, Glenn Creek area (65°18'N, 142°05'W). 7 – 18 August 2001. The NPS cabin at Glenn Creek was used as a base of operations to sample a variety of habitats in the area, including sites near Logan and Rock creeks.

Yukon River, Lower Kandik River $(65^{\circ}22.4^{\circ}N, 142^{\circ}30.4^{\circ}W)$. 13 - 18 August 2001. The immediate area at the mouth of the Kandik River was sampled from the NPS cabin located here.

Field Studies

Our collecting strategy was designed to maximize the number and diversity of samples by using a variety of methods in available habitats. While particular effort was made to sample rare or undocumented small mammals, the sampling methods used also allowed us to evaluate the occurrence and relative abundance of the more common species.

Diversity of captured specimens was maximized by utilizing a variety of trap types, including snap traps (Museum Specials, rat traps) and live traps (44 oz. plastic drinking cups buried as pitfall traps, Sherman live traps, buried 5-gal. Buckets). Marmots, arctic ground squirrels, red squirrels, collared pikas, and porcupines were usually collected with shotgun or rifle.

Traplines for shrews and voles were set in the range of available habitats and ecotones in each study location. Traplines typically consisted of 20 or more trap stations per line, with stations spaced 8-10 m apart. At each station, 2 snap traps or 1 snap trap and 1 pitfall trap were typically set within 2 m of each station point. The snap traps were baited with a mixture of rolled oats and peanut butter; pitfall traps were buried flush with the ground and left unbaited. Traps were usually set in the late afternoon and checked the following morning. Productive lines were usually kept in operation for 2 or more nights.

Specimen Processing

All animals sampled were preserved as a scientific specimen. Specimens were prepared as skeletal preparations or as whole bodied alcoholics. A small number of dried study skins were also prepared. Each crew carried tank of liquid nitrogen in the field to preserve tissues (heart, liver, kidney, spleen, and lung) and embryos. These frozen specimens were transferred to ultralow temperature freezers at UAM and stored at -70° C. We preserved ectoparasites, endoparasites and feces samples from many of the mammals collected. These exceptional data sets will be used to address epidemiological, coevolutionary, taxonomic, and biogeographic questions. Intestinal tracts from shrews were also preserved. Protocols followed to rigorously document and preserve specimens are included in the appendix.

All mammal specimens from this study have been accessioned into the mammal collection at UAM and are in process of curation and data entry. Skeletal material from a subsample of

Microtus voles are still in process of being cleaned and positively identified (as either *M. pennsylvanicus* or *M. oeconomus*). The samples of endoparasite are now at the US National Parasite Lab in Beltsville, MD, and ectoparasites are at Idaho State University. Feces samples (for Coccidia) are already under study at the University of New Mexico, and lung samples (for Hanta virus) are being examined by colleagues in Finland.

Results and Discussion

Inventory Results

The specific products of this inventory include a large collection of well-prepared, well-documented, and diverse preparations of mammal specimens and associated materials (tissues, parasites, fecal samples, digestive tracts). A total of 1891 small mammal specimens (excluding embryos) comprising 17 species was archived from 10 general collecting localities in YUCH between 16 July-19 August 2001 (Table 1).

The cinereus shrew and the northern red-backed vole were the most frequently captured species, comprising 57% of all specimens collected (Figure 2). Meadow voles ranked 3rd (17% of captures). Fourteen additional species, in descending order of specimen abundance, were the tundra vole, tundra shrew, northern bog lemming, collared pika, montane shrew, red squirrel, pygmy shrew, long-tailed vole, arctic ground squirrel, tiny shrew, brown lemming, hoary marmot, North American porcupine, and taiga vole.

We found most species present across the range of elevations sampled (Figure 5). Four species were restricted to alpine sites (hoary marmot; arctic ground squirrel, but see species account; collared pika; brown lemming), while three species were essentially confined to the lower elevations (pygmy shrew, tiny shrew, red squirrel).

The findings from this study, when combined with specimen information gathered from our review of holdings in other major collections, bring the total number of documented small mammal species to 21 (out of a probable 24), or 88% of the small mammal species that most likely inhabit the Preserve (Table 2).

The discovery of tiny shrews (*Sorex yukonicus*) constitutes a new species for the Preserve and a major range extension for the species.

Species Accounts

The following accounts summarize information on each species known or suspected to occur in YUCH. Species observed but not collected are indicated with an asterisk (*). Species that have been reported or might possibly occur in the Preserve but have not been adequately documented are marked with a dagger (†). Detailed data on all specimens will be available in the museum database and accessible on its website once the time consuming process of curation is completed sometime in 2002.

Order INSECTIVORA—Shrews

Family Soricidae

Sorex cinereus, cinereus shrew

We captured more cinereus shrews than any other small mammal species. Only red-backed voles rivaled them in number and ubiquity across the landscape. This shrew occurred in all major habitats and at most elevations sampled. They were numerous at lower elevations particularly in open, grassy and moist situations. Pitfall traps accounted for most shrew captures. The cinereus shrew is the dominant shrew in many communities throughout central Alaska. Their abundance

and ecological flexibility may be responsible, at least partially, for the general scarcity of other shrew species (Wrigley et al. 1979).

Sorex hoyi, pygmy shrew

We captured 24 pygmy shrews at various lowland and subalpine sites along and north of the Yukon River. No pygmy shrews were captured in the mountains of the upper Charley River.

The only previous report of this species from within the Preserve is a single specimen taken by Clough (1976) from near the mouth of the Kandik River. Several pygmy shrew skulls were identified in a collection of small mammal skeletal material recently donated to UAM by Brad Shults, NPS-Fairbanks. They were taken the Logan Creek area in the early 1990s.

Sorex monticolus, montane shrew

Nowhere abundant, a total of 39 montane shrews were sampled at various elevations in a variety of habitats. Osgood (1900) found this species most prevalent in the mountains and documented the first and only specimen taken within the Preserve.

†Sorex palustris, water shrew

No water shrews were seen or collected during this study; however, the recent discovery of water shrews on a tributary of the Yukon River at Big Windy Hot Springs (Cook et al. 1997), approximately 27 km from the westernmost boundary of YUCH, and along the Klondike River in neighboring Yukon Territory (Jarrell 1986), suggest their probable occurrence in the Preserve.

Sorex tundrensis, tundra shrew

Tundra shrews were a distant second in total shrew captures, comprising 96 individuals from a variety of sites and elevations. Most came from traplines set below treeline in open forests, dwarf tree woodlands, and mesic meadows.

Osgood (1900) collected 5 specimens of tundra shrews within the present boundaries of the Preserve. A *S. tundrensis* skull was included in material provided to UAM by Brad Shults.

Sorex yukonicus, tiny shrew

The tiny shrew (Figure 3) is a new species to YUCH and its capture constitutes a major range extension. Prior to this inventory, only 12 specimens of *S. yukonicus* were known to science (Dokuchaev 1997; pers. com. 2001). All are from Alaska, with the closest records to the Preserve from the upper Susitna River basin and far down the Yukon River near Ruby.

Despite considerable effort, only 10 tiny shrews were captured, and only in the vicinity of Kathul Mountain. Nine of the 10 were taken in pitfall traps set in a diversity of forest, dwarf woodland, and tall scrub habitats at elevations ranging from river bottom to just beyond the edge of trees (Figure 4).

Order CHIROPTERA—Bats

Family **Vespertilionidae**

†Myotis lucifugus, little brown bat

No bats were seen or reported. Unidentified bats, most likely of this species, have reportedly been sighted downriver at Fort Yukon as well as upriver near Dawson (Parker et al. 1997). This bat may a sporadic visitor to the Preserve.

Order **CARNIVORA**—Carnivores

Family Canidae

Canis latrans, coyote

No coyotes or their sign was observed or reported, although it is possible individuals may occasionally visit the area from extant populations further south in the Tanana Valley or from neighboring Yukon Territory. No preserved specimens from this area are known to exist. Coyotes are said to have once occurred here, arriving as newcomers to the state first near Eagle, sometime around the early 1920s (Rearden 1981) and reaching peak numbers about 1940 (Dufresne 1946).

*Canis lupus, wolf

We frequently encountered wolf sign during our study. A cow moose with calf was observed seeking water refuge from an interested wolf near Glenn Creek.

Vulpes vulpes, red fox

Fox tracks were noted along the banks of the Yukon River.

Family Felidae

*Lynx canadensis, Canada lynx

Individual lynxes were encountered at Coal and Glenn creeks, and one was seen swimming across the Yukon River below Woodchopper.

†Puma concolor, Puma

A local resident of Eagle told of seeing (in the past year) the tracks of a puma along the Yukon River below Eagle. The validity of this report is very questionable but not impossible given the apparent northwestward expansion of this species' range in recent years (MacDonald and Cook 1996).

Family Mustelidae

*Gulo gulo, wolverine

A lone wolverine was seen in the mountains of the upper Charley River.

†Lontra canadensis, northern river otter

No river otters or their sign was observed or reported during our time in the area. This species is a probable uncommon resident of the upper Yukon River Valley. UAM has a number of river otter specimens from near Eagle and Circle.

†Martes americana, American marten

The marten is a common resident of the Preserve. It is the primary furbearer species still being commercially harvested in the Preserve. Martens are primarily nocturnal, so it was not surprising that none were encountered during our stay in the area.

*Mustela erminea, ermine

A single ermine was observed near Slaven Cabin. Osgood (1900) secured ermine specimens from this area. More ermine specimens are needed to clarify the relationship and geographic extent of two distinct populations that come in contact in this area of the state (Eger 1990, Fleming and Cook *accepted*).

†Mustela nivalis. least weasel

No least weasels were collected or observed, and no specimens are known from this area. The status and distribution of this species in the region needs to be clarified. The species is generally uncommon and sparsely distributed throughout much of its holarctic range. It occupies a wide

variety of forest and tundra habitats, but favors meadows, marshes, and riparian situations where its small rodent prey are found in abundance (Banfield 1974).

†Mustela vison, American mink

No sightings or sign of this resident species were noted during the course of our field work. No specimens are known from the Preserve.

Family Ursidae

*Ursus americanus, American black bear

We frequently encountered black bears and their sign within the forest zone of the Preserve. Several times mischievous black bears caused havoc to traplines in the Slaven Cabin area.

*Ursus arctos, brown bear

Lone brown bears were seen on two occasions in the mountains of the upper Charley River. One was also observed near Squaw Mountain and another, a young animal, was seen running below the steep banks of the Yukon River near the westernmost boundary of the Preserve.

Order ARTIODACTYLA—Ungulates

Family Cervidae

*Alces alces, moose

Our field crews encountered moose and/or their sign throughout the study area.

*Rangifer tarandus, caribou

Various-sized groups of caribou were frequently encountered in the mountains of the upper Charley River. We found the shed antlers of caribou near Squaw and Kathul mountains, but no animals were seen while we were in these areas.

Family **Bovidae**

*Ovis dalli, Dall's sheep

Dall's sheep were noted in the upper Charley River and at Squaw Mountain near the US/Canada border. One sheep seen at Squaw Mountain was distinctly brown-backed.

Order **RODENTIA**—Rodents

Family **Sciuridae**

†Glaucomys sabrinus, northern flying squirrel

We failed to secure any specimens of flying squirrels during the course of our fieldwork. This species undoubtedly occurs in forested areas of the region and a survey of marten trappers active in the Preserve would confirm their presence and provide valuable specimens for study. The type specimen for the subspecies *G. s. yukonensis* was collected near the Alaska-Canada border above Eagle (Osgood 1900).

Marmota caligata, hoary marmot

Marmots were found in rocky situations high in the mountains of the upper Charley River. We obtained 4 specimens in the upper Crescent Creek area. No marmots were seen or heard near Squaw Mountain north of the Yukon River; however, they are known to occur in the Ogilvie Mountains in adjacent Yukon Territory (Osgood 1900, Youngman 1975)

†Marmota monax, woodchuck

The occurrence of woodchucks anywhere in east-central Alaska away from the Tanana Valley remains speculative. The type specimen of the subspecies, *M. m. ochracea*, is from the head of

Fortymile River (Swarth 1911). UAM has a woodchuck specimen from 50 miles up the Salcha River. The species occurs in the Dawson area of Yukon Territory (Youngman 1975).

Spermophilus parryii, arctic ground squirrel

Two widely separate and morphologically distinct populations of arctic ground squirrels occur within the boundaries of the Preserve. One is restricted to alpine areas of the Ogilvies where these mountains extend into the NE corner of the Preserve from neighboring Canada. We found ground squirrels numerous in alpine tundra habitats in the vicinity of Squaw Mountain, but none farther west near Kathul Mountain or in the mountains to the south in the upper Charley River country. Squaw Mountain squirrels are probably referable to the subspecies *S. p. plesius* (Youngman 1975, Hall 1981).

A second population, referable to *S. p. osgoodi* from lowland areas of the Yukon Flats region, has been found along the banks of Yukon River at the extreme northwestern boundary of the Preserve (Osgood 1900, USNM, UAM).

The taxonomy of this holarctic species has been contentious and needs formal revision. Additional samples of Yukon Valley squirrels would not only help clarify the taxonomic and phylogeographic relationships of the two disjunct populations in the Preserve, but could also contribute significantly to our understanding of the species as a whole.

†Tamias minimus, least chipmunk

Least chipmunks are known to occur in the Yukon Valley as far west as Dawson in Yukon Territory (Youngman 1975). Although we found no evidence of their range extending farther downriver into Alaska or the Preserve, continued effort should be expended to confirm the absence of this species..

Tamiasciurus hudsonicus, red squirrel

Red squirrels were common in the spruce and mixed forests of the Preserve. A single red squirrel was heard calling from a stand of spruce at the very edge of treeline (1019 m elevation) near Squaw Mountain.

Family Castoridae

*Castor canadensis, American beaver

We frequently encountered beavers and signs of their activities while working along the Yukon River and its tributaries. The mosaic of wetland habitats provided by the industry of this keystone species proved very productive for small mammals, especially shrews, meadow voles and bog lemmings.

Family **Dipodidae**

†Zapus hudsonius, meadow jumping mouse

No jumping mice were collected despite focussed trapping efforts in the grassy-tall shrub habitats of riparian areas they are known to prefer elsewhere. The closest records of this species are upriver near Dawson (Youngman 1975) and downriver near Rampart (UAM).

Family Muridae

Clethrionomys rutilus, northern red-backed vole

Red-backed voles occurred at all localities and in all major habitats. The species was most frequently captured in forest and scrub communities and least often in alpine tundra and wet meadows. Red-backed voles, together with cinereus shrews, were the dominant mammal species of the region.

†Dicrostonyx groenlandicus, collared lemming

No collared lemmings were collected. Small numbers of collared lemmings, described as a distinct and isolated subspecies *D. torquatus nunatakensis* by Youngman (1975), have been found in rocky alpine tundra in the Ogilvie Mountains NE of Dawson in Yukon Territory. The current status and distribution of this taxon is unknown.

Lemmus trimucronatus, brown lemming

Only 5 brown lemmings were sampled during our inventory. All were found at or above treeline mostly in wet herbaceous and low scrub situations. Previous studies in YUCH (Osgood 1900, Brad Schults collection) secured small numbers of brown lemmings from sites at lower elevations, including the type specimen for *L. t. yukonensis* from the mouth of Kandik River (Osgood 1900).

Dr. Vadim Fedorov, UAM, has now sequenced 915 base pairs of mtDNA cytochrome *b* of the *Lemmus* specimens collected this past summer. His findings suggest that two divergent (6%) phylogeographic groups come in contact within the boundaries of the Preserve. The lemming from Squaw Mountain (and also one from Chisana in Wrangell-St. Elias National Park and Preserve) had a Beringian (i.e., Alaska, eastern Siberia) haplotype while animals from Kathul and Copper mountains (also Braye Lakes, WRST) had haplotypes of a more Eastern group (Canadian arctic east of the Mackenzie Delta and, interestingly, an animal from the upper Susitna River in southcentral Alaska). More specimens are needed from throughout the Preserve to document the extent of these lineages.

Microtus longicaudus, long-tailed vole

Small numbers of long-tailed voles were collected at 7 of 10 localities. Most were restricted to rocky situations on dry grassy sites, from steep south-facing slopes bordering the Yukon River (Figure 8) to talus areas high up in the mountains.

†Microtus miurus, singing vole

This semi-colonial vole occurs in a variety of tundra and taiga-tundra habitats on well-drained sites, sometimes along streambanks, near or above treeline. This species inhabits many of the mountainous areas of Alaska, but is thought not to occur today in the Yukon-Tanana uplands (e.g., Kurtén and Anderson). Clough (1976), however, collected a singing vole on Mount Kathyrn and another on Twin Mountains during a 1975 survey, and Youngman (1975) reported them from the Ogilvie Mountains in neighboring Yukon Territory.

Despite considerable effort to secure additional specimens of this species, we failed to find any evidence of their presence in either the mountains of the upper Charley River (including Mount Kathyrn) or at Squaw Mountain at the edge of the Ogilvies.

Perhaps populations were critically low and patchy during the time of our survey or, less likely, Clough (1976) was mistaken in his identifications. [At our request, Clough's Mt. Kathyrn specimen was re-examined by Dr. Robert Hoffmann at the USNM and confirmed as M. miurus.]

Microtus oeconomus, tundra vole

Tundra voles were found at 8 of the 10 localities surveyed. They were generally restricted to open, non-forested sites, most often in moist grass-sedge/scrub habitats in the mountains. At lower elevations, the numerically-dominant and ecologically-similar meadow vole may have helped relegate tundra voles to more marginal sites such as disturbed scrub/forb thickets along the Yukon River.

Microtus pennsylvanicus, meadow vole

The meadow vole was the third most frequently captured species in 2001. They were most abundant in grassy wetland habitats at lower elevations. This vole was also found in wet grassy situations along drainageways and in swales farther up in the mountains.

Microtus xanthognathus, taiga vole

A single taiga vole captured in subalpine spruce woodland near Squaw Mountain was our only encounter with this species during the study. Efforts to locate active colonies near Glenn, Logan, and Rock creeks were unsuccessful. This species inhabits fire-successional and riparian, boreal, sphagnum forest habitats near streams and other moist areas. Their populations are known to be ephemeral, patchy, and unpredictable (Wolf 1999). Osgood (1900) found *M. xanthognathus* sparingly distributed along the Yukon River from Eagle to the vicinity of Circle, and secured a small series of specimens for the USNM from near the mouth of the Kandik River and 32 km upriver from Circle. Shults et al. (1993) reported the total capture of 72 taiga voles in 1991 and 1992 on burned and unburned sites near Logan Creek.

†Ondatra zibethicus, muskrat

Muskrats were not encountered during the course of our field work in the Yukon Valley. Osgood (1900) provided a single specimen from 24 km below Eagle.

†Peromyscus maniculatus, deer mouse

We found no evidence of deer mice occurring in the area, although a report of "mice" inhabiting human dwellings in Eagle should be investigated. The species has been found upriver in Dawson (Youngman 1975).

Synaptomys borealis, northern bog lemming

Northern bog lemmings were relatively common and widespread in 2001. Most were captured in wet grassy-scrubby meadows and open tussocky bogs.

Family Erethizontidae

Erethizon dorsatum, North American porcupine

Porcupines were rare but widespread. Single animals were collected above treeline at upper Crescent Creek and near Mount Kathryn. Another was secured and a second one seen in the Yukon Valley near Slaven Cabin.

Order LAGOMORPHA—Pikas and Hares

Family Ochotonidae

Ochotona collaris, collared pika

Pikas were common in scree slopes above treeline. Samples were taken at upper Crescent Creek, Mount Sorenson, Mount Kathyrn, and Squaw Mountain. No pikas were found above treeline near Kathul Mountain.

Family **Leporidae**

*Lepus americanus, snowshoe hare

Snowshoes hares appeared to be at a low stage of their population cycle during the time of our visit. We saw single animals at the edge of forest near Kathul Mountain and in riparian scrub along Coal Creek. None were sampled.

Habitat Affinities

Habitats of small mammals are usually related to, and often defined by, the association of plants at that site. Under the influences of the topography, soils, climate conditions, and other ecological factors, plants tend to fall into distinct groups referred to as vegetative communities, associations, or types. A mammal species usually is associated with particular groups of plant communities (at various macro- to micro-scales of resolution). Some species are restricted to one such community, some to two or more. The degree of species' dominance in and range across various types may be most related to population levels that fluctuate dramatically from year to year and even season to season. These fluctuations are particularly acute at high latitudes. Interspecific interaction and density may be major influencing variables, particularly among similar species (e.g., *Microtus pennsylvanicus* and *M. oeconomus*, or among *Sorex* shrews).

Besides vegetation, other features and factors may influence a species' distribution, including topography, soil types, snow cover, availability of food, and/or the presence of other important features such as water bodies, rocks, and ground litter. Each species also has its own biogeographic and evolutionary history and ability to tolerate different situations and circumstances.

Shrews, voles and lemmings were unevenly distributed over the range of elevations (Figure 5) and vegetation types sampled (Table 3). Patterns of habitat occupancy among species indicated that the two most common species, cinereus shrews and red-backed voles, are habitat generalists, occurring relatively evenly across a broad range of habitat types in the study area (Figure 6a,b). Red-backed voles were most abundant in forest and scrub habitats within the forested zone. A small number of red-backed voles were captured in scrubby situations above treeline. The local distribution of this common species may have been tied most closely to the presence of overhead cover, especially woody plant cover. Tall tussocks may also have served as overhead protection.

The general habitat requirements of shrews is probably most related to invertebrate abundance and physical conditions such as temperature and moisture (Nagorsen 1996). We found cinereus shrews occurring in most major habitats but especially in riparian areas, wetlands, moist forests and tussock bogs and woodlands at the lower elevations. Tundra shrews, pygmy shrews, and tiny shrews occupied sites at or below treeline in a variety of habitats, particularly ones having adequate ground cover in the form of leaves, stumps, decayed logs, and vegetation. Montane shrews were found in similar situations within the taiga as well as in the alpine tundra. Tundra shrews and montane shrews (and perhaps tiny shrews) may have favored slightly drier sites than either cinereus shrews or pygmy shrews. All 5 species were found coexisting in an open dwarf tree scrub/tussocky site at the edge of treeline E of Kathul Mountain (Figure 4, top left photo).

In contrast to the ecological flexibility displayed by red-backed voles, voles of the genus *Microtus* and lemmings demonstrated stronger habitat specificity, as evidenced by their restriction to herbaceous and herbaceous/scrub vegetation types in both taiga and tundra environments. The few sites where brown lemmings were captured ranged from wet grass-sedge openings within subalpine woodland and scrub (Figure 7, top) to dry alpine scree. Meadow voles and bog lemmings were relatively numerous during the time of this study and were generally restricted to the wetter graminoid meadows and boggy sites within the taiga zone (Figure 7, bottom). The limited number of long-tailed voles captured suggested their preference for drier grassy sites in rocky situations at various elevations (Figure 8). Tundra voles occurred in a variety of open herbaceous/scrub sites above and below treeline. Tundra voles may have a greater tolerance to different unforested habitats than meadow voles.

Overall, small mammals were most diverse (species richness) at the transition zone between taiga and alpine tundra, and most abundant (and also diverse) in the mosaic of bottomland habitats of the Yukon Valley.

Single-season surveys provide only a limited assessment of small mammal habitat. A more accurate and meaningful assessment will require a focused ecological approach that tracks dramatic population fluctuations among species over an extended period of time.

Summary and Significance

Findings of this inventory confirm the importance of YUCH as home to a rich assemblage of northern mammals. Of the Preserve's approximately 40 resident species of mammals, over half are small mammals. Some 88% of the small mammals suspected to occur in YUCH are now documented with specimens. From tissue samples collected during this study, mammalian diversity of the Preserve has been expanded by the discovery of two co-occurring lineages of brown lemmings. Similar studies of other taxa (e.g., ermine, arctic ground squirrels) could provide additional support to this initial hypothesis that YUCH is an important zone of contact for a number of phylogeographic groups of mammals.

Patterns of relative abundance and general habitat occupancy among 17 species sampled in the Preserve were similar to those reported from other small mammal studies in Central Alaska (e.g., Buckley and Libby 1957, Pruitt 1968, West 1979, MacDonald 1980, Buskirk and MacDonald 1984). Three species—an insectivore (*Sorex cinereus*), a generalized fruit-seed-leaf feeder (*Clethrionomys rutilus*), and a specialized graminoid grazer (*Microtus pennsylvanicus*)—dominated the small mammal community of the study area, accounting for nearly 80% of all shrew and small rodent captures.

Our discovery of the tiny shrew, *Sorex yukonicus*, perhaps the rarest and poorest known mammal in North America, was an anticipated surprise. The 10 tiny shrews sampled in the Preserve significantly expanded the known range of the species and nearly doubled the number of specimens known to science.

Several likely-occurring species remain undocumented from the Preserve. These include species such as the water shrew, *Sorex palustris*, least weasel, *Mustela nivalis*, Northern flying squirrel, *Glaucomys sabrinus*, and perhaps meadow jumping mouse, *Zapus hudsonius*.

The most significant and valuable product of this inventory is the large collection of well-documented and diverse preparations of scientific specimens.

Why specimens? As elucidated by Reynolds et al. (1996), voucher specimens and corresponding data assembled during field surveys of mammals, particularly the smaller species that are difficult to identify (e.g., shrews, *Microtus* voles) and often poorly known (most of Alaska small mammals), are critical for accurate identification of the animals studied and for verification of the data gathered and reported as resulting from the investigation.

Voucher specimens and their associated materials are critical for a wide array of studies such as taxonomic revisions, biogeographic and evolutionary studies, parasitology, and epidemiology.

Voucher specimens provide critical historical baseline for assessment of change caused by natural or human perturbations. As they represent historical populations, the value of large series of specimens increases through time, particularly as the diversity of many localities is degraded. With PCR (polymerase chain reaction) and other innovations in the study of DNA, we now can examine and monitor genetic variation in populations of animals that were collected during different time periods; thus providing a more rigorous view of temporal genetic variation and population structure. For example, known contact zones between taxa can now be reanalyzed for temporal stability (but only if specimens from the contact zone were collected at regular intervals). Because of the dynamic geologic history of Alaska and the role that glaciers played in the distribution of organisms, these kinds of studies are essential to documenting and managing biodiversity.

Without the preservation of specimens, inventories such as this one would have extremely limited value (either short-term or long-term). As pointed out by Reynolds et al. (1996), it is essential that agencies requiring and supporting biodiversity assessments recognize the critical need for vouchers and provide support in both field and museum budgets for their preservation and maintenance.

While the importance of museum specimens should be generally recognized and their preparation considered essential to good science, for many the question remains: Why so many? Some perspectives:

- Alaska mammalogy is still in the early exploration phase. Many areas are poorly known
 and inadequately represented in systematic collections. YUCH is no exception. Prior to this
 study, preserved specimens from this area were essentially limited to samples of skin and
 skeletal materials obtained by W. H. Osgood and assistants when they passed through here
 in the summer of 1899 and again in 1903. More than 70 years later, G. C. Clough (1976)
 added 30 additional skin/skull specimens of 11 small mammal species to the USNM
 collections.
- Small numbers of specimens will not adequately sample the variation in and between populations. Many studies require large samples of well-preserved (and diverse) study material to account for age, sex, geographic, and/or individual variation. Taxonomic studies based on skull morphology may require undamaged material from 20 or more adult individuals of each sex per locality. For perspective, of the 52 red-backed voles sampled at the mouth of the Kandik River in this study only 12 (6 males, 6 females) are clearly adult and therefor useful for making morphological comparisons.
- Many of the shrews and small rodents are difficult or impossible to tell apart except by specimen. Close examination of tooth pattern and comparison of body measurements and other characters is necessary to tell most of Alaska's shrews apart. *Microtus* voles can also be especially difficult to differentiate. *M. oeconomus* and *M. pennsylvanicus*, in particular, are so similar that positive identification can only be made by a magnified look at their molars (either by surgery in the field or later on in the lab after the skulls are cleaned).
- Many captures of the most common and widespread species may be necessary in order to document the rare and uncommon. In this survey, several thousand trapnights were needed to find the 10 tiny shrews amidst the many, many cinereus shrews.
- As pointed out in Reynolds et al. (1996), the number of animals removed from a population
 has no biological significance unless it is related to the total number of animals in the
 population and their rate of replacement. Alaska's small mammals are short-lived and
 prolific, with reproductive potentials more than sufficient to accommodate low levels of
 removal found in our inventory project. As added perspective, the total mass of all shrews,
 voles and lemmings sampled in YUCH in 2001 was roughly equivalent to one averagesized beaver.

Recommendations for Future Inventory and Monitoring Efforts

- 1. Continued efforts are needed to document the occurrence of several mammal species in YUCH.
 - Water shrews are most likely present within the boundaries of the Preserve but perhaps only at special sites and situations. Areas with year-round open water such as the geothermal springs at Big Windy Hot Springs just west of the Preserve should be identified and sampled for this semi-aquatic shrew.

- The meadow jumping mouse may also inhabit particular riparian habitats in the Yukon Valley and should be surveyed for.
- Occasionally little brown bats may pass through the region, particularly late in the summer and early autumn. Voucher specimens will be required to validate identities.
- Reports of mice in homes at Eagle should be followed up to determine if these may be deer mice or non-native house mice (*Mus musculus*).
- The status of the least weasel, ermine, and flying squirrel need to be clarified. Help from commercial trappers should be sought to help provide adequate samples of these species.
- 2. YUCH has been identified as a general area of contact for two distinct phylogeographic groups of brown lemmings and ermines. Populations of other mammal species may display similar patterns and should be studied using modern techniques with adequate samples. Specimens that include frozen tissues from lowland populations of arctic ground squirrel (*Spermophilus parryii*) at the western edge of the Preserve need to be sampled and compared with the good series taken near Squaw Mountain.
- 3. Further inventory work is needed throughout the Ogilvie Mountains portion of the Preserve. Efforts to secure the necessary permissions and permits from Doyon Corporation to access the most promising sites in this area need to be continued. Several species still need to be documented from here, including hoary marmot (*Marmota caligata*), singing vole (*Microtus miurus*), and possibly collared lemming (*Dicrostonyx groenlandicus*). Specimen material from these and other species will be extremely valuable in future taxonomic and phylogeographic research projects. Dall's sheep (*Ovis dalli*) from this and other populations in the Preserve could also be an important species in a phylogeographic analysis of the region and efforts should be made to secure an adequate sampling of tissue and voucher material.
- 4. We contend that long-term monitoring on biotic change is best accomplished by preserving materials from populations sampled periodically over time. Specimen-based monitoring of northern small mammal populations has been ongoing in Scandinavia for many decades. Dr. Heikki Henttonen, esteemed colleague from Finland and participant in this study, has been principal investigator of such efforts for several decades. We encourage NPS to work closely with individuals such as Dr. Henttonen to develop a rigorous monitoring program in Alaska.

Acknowledgments

A project of this kind involved the hard work and diverse talents of many. Our thanks to Dr. Heikki Hentonnen (Finland), Dr. Jukka Niemimaa (Finland), Anatoli Lazutkin (Russia), Anna Goropashnaja (Russia), Andrew Lynch (University of New Mexico), Sarah Runck (University of Alaska), Nena MacDonald (New Mexico), Lourdes Barrelli (University of Alaska), Kabai Gamblin (Idaho State University), Nikki Guldager (NPS-Fairbanks), John Burch (NPS-Fairbanks), Sara Wesser (NPS-Anchorage), Shelli Swanson (NPS-Fairbanks), Marsha Henderson (NPS-Fairbanks), Sterling Holbrook (helicopter support), Rick Swisher (Quicksilver Air), Brandy Jacobsen (UAM), Amy Runck (UAM), John Bender II (UAM), Eric Waltari (UAM), Eric Tomasik (Idaho State University), Dr. Vadim Fedorov (UAM), Dr. Gordon Jarrell (UAM), and others not listed above.

Cover drawing of *Microtus pennsylvanicus* is from Bailey (1900).

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Table 2. Species of small mammals that have been documented in the Yukon-Charley Rivers National Preserve (\odot = voucher specimen; \bigcirc = observed but not documented by specimens).

SPECIES	This Study	Other Studies	Notes
INSECTIVORA – Shrews			
Family Soricidae			
Sorex cinereus, cinereus shrew	•	•	
S. hoyi, pygmy shrew	0	•	
S. monticolus, montane shrew	0	•	
S. palustris, water shrew			Known to occur just west of Preserve
S. tundrensis, tundra shrew	•	0	
S. yukonicus, tiny shrew	•		New species for Preserve
CARNIVORA – Carnivores			
Family Mustelidae			
Mustela erminea, ermine	0	0	USMN specimens
M. nivalis, least weasel			"Upper Yukon" (USNM)
RODENTIA - Rodents			
Family Sciuridae			
Glaucomys sabrinus, N. flying squirrel			Specimens upriver from Eagle just inside Yukon Territory (USNM)
Marmota caligata, hoary marmot	•	0	made Taken Territory (Cortin)
Spermophilus parryii, arctic gnd. squirrel	0	0	
Tamiasciurus hudsonicus, red squirrel	0	0	
Family Muridae			
Clethrionomys rutilus, N. red-backed vole	•	•	
Lemmus trimucronatus, brown lemming	•	•	Type specimen for L. t. yukonensis from "Charley Creek (= Kandik R.)
Microtus longicaudus, long-tailed vole	0	· ·	Hom Charley Creek (Randik R.)
M. miurus, singing vole		<u> </u>	Specimen in USNM by Clough (1976) verified as <i>M. miurus</i> by Dr. Robert Hoffmann
M. oeconomus, tundra vole	•	0	
M. pennsylvanicus, meadow vole	•	0	
M. xanthognathus, taiga vole	•	•	
Ondatra zibethicus, muskrat		•	USNM specimen
Synaptomys borealis, N. bog lemming	0	0	
Family Erethizontidae			
Erethizon dorsatum, N.A. porcupine	0		
LAGOMORPHA – Pikas & Hares			
Family Ochotonidae			
Ochotona collaris, collared pika	0	•	
Family Leporidae			
Lepus americanus, snowshoe hare	0	0	USNM specimen

POSSIBLE ADDITIONAL SPECIES: Myotis lucifugus, little brown bat, Marmota monax, woodchuck, and Zapus hudsonius, meadow jumping mouse, may yet be found in YUCH. Peromyscus maniculatus, deer mouse, and Tamias minimus, least chipmunk, are known from the Dawson area in Yukon Territory. Dicrostonyx groenlandicus, collared lemming, has been taken in the Ogilvie Mountains in neighboring Yukon Territory.

Table 3. Number of shrew and small rodent captures and trapping effort in vegetation types (Levels I-III of Viereck et al., 1992) sampled at 8 of 10 general localities in the Yukon-Charley Rivers National Preserve, Alaska, in July and August, 2001.

	LEVEL I				FO	RE	ST							•	SCF	RUB	i					ŀ	IERI	BAC	EC	วบร		П
	II		EDI LEA			ROA EAF		М	IXE	D		VAF		TA	LL	LO	W	D۱	VAF	₹F	_	RAI NOI		F	OR	В	BR OI	
	HI	CLOSED	OPEN	WOODLAND	CLOSED	OPEN	WOODLAND	CLOSED	OPEN	WOODLAND	CLOSED	OPEN	WOODLAND	CLOSED	OPEN	CLOSED	OPEN	Dryas	Ericaceous	Willow	DRY	MESIC	LEM	DRY	MESIC	WET	Mosses	Líchens
	INSECTIVORA - Shrews		Ĺ																									
	Family Soricidae						<u> </u>																					
	Sorex cinereus, cinereus shrew	6	54	12				41					46	9	61		15	1		2	18	31	70	3				_
	S. hoyi, pygmy shrew		1	1				3					5		1								2	2			1	_
	S. monticolus, montane shrew			В				1					3	2	3		8				2	3	1					_
	S. tundrensis, tundra shrew		12					5					11	1	3		4					9						
ES	S. yukonicus, tiny shrew		3			_		1					1		1	-												\dashv
SPECIE	RODENTIA - Rodents																											彐
윤	Family Muridae	<u> </u>																										_
	Clethrionomys rutilus, N. red-back. vole	18	24	9				35					42	8	60	1	19				27	18	47	10				2
MAMMAL	Lemmus trimucronatus, brown lemming	<u> </u>	1				L				L		1		1		1_							1				_
₽	Microtus longicaudus, long-tailed vole	L		1													11				5			2				_
¥	M. oeconomus, tundra vole		1					1					9	6	9		12					28		1				
≥	M. pennsylvanicus, meadow vole	1	1		!			1					3	2	41		7				2	18	116					_
	Synaptomys borealis, N. bog lemming	1	3	1	L			1		L	Щ		3		7		8						18		\sqcup			_
		<u> </u>	<u> </u>																									_
																									4	_		
	TRAP NIGHTS	180	996	700				928					1132	345	1927	108	825			72	340	1190	1305	746				546

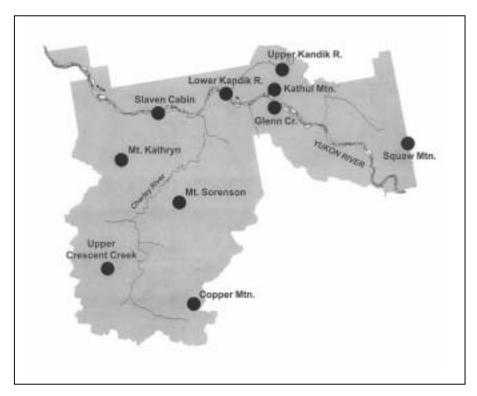


Figure 1. General localities in the Yukon-Charley Rivers National Preserve, Alaska, sampled for small mammals in July and August 2001.

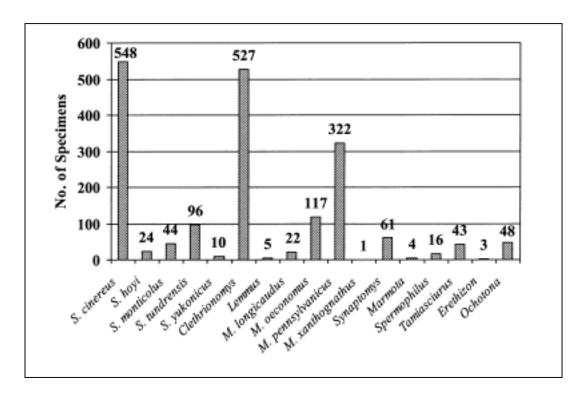


Figure 2. Total number of specimens of 17 species of small mammals sampled in the Yukon-Charley Rivers National Preserve, Alaska, in July and August 2001.



Figure 3. Tiny shrew (*Sorex yukonicus*) captured E. of Kathul Mountain, Yukon-Charley Rivers National Preserve, Alaska, on 26 July 2001. Approximately actual size (external measurements in mm: 74-22-9-xx=2.3 g, male; AF 52116).







Figure 4. Some sites where tiny shrews (*Sorex yukonicus*) were captured E. of Kathul Mtn. (approx. 950 m elevation), Yukon-Charley Rivers National Preserve, Alaska, July 2001.

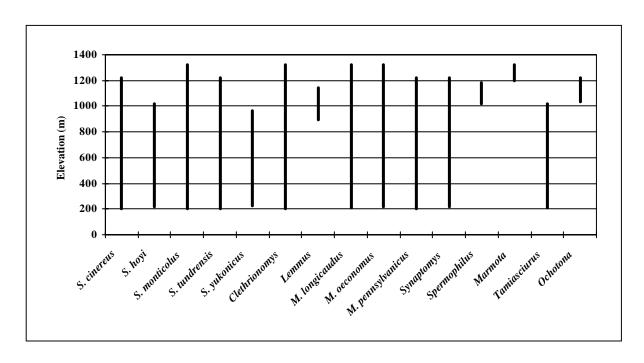


Figure 5. Range of elevations where small mammal species were captured in July and August, 2001, Yukon-Charley Rivers National Preserve, Alaska.

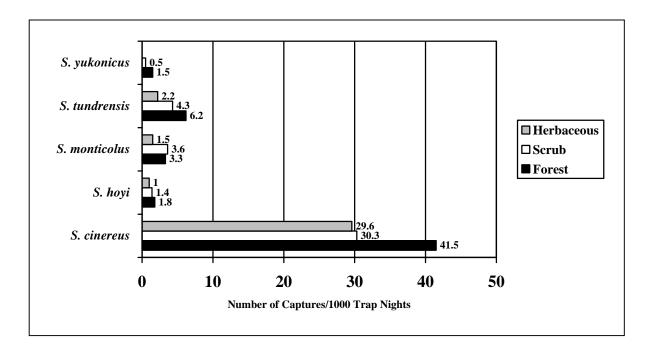


Figure 6a. Relative abundance (specimens/1000 trapnights) of shrews in major vegetation types, Yukon-Charley Rivers National Preserve, Alaska, July-August 2001.

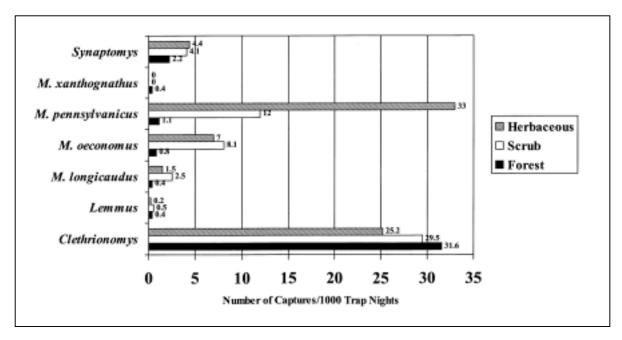


Figure 6b. Relative abundance (captures/1000 trap nights) of voles and lemmings in major vegetation types, Yukon-Charley Rivers National Preserve, Alaska, July-August 2001.





Figure 7. Capture site of brown lemming (top photo); meadow voles, bog lemmings, and cinereus and pygmy shrews occurred around the fringe of this marshy pond (bottom photo).

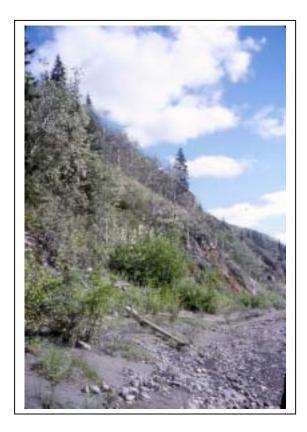


Figure 8. Several long-tailed voles were captured near rocks on this dry south-facing slope along the Yukon River.

Appendix

SPECIFIC COLLECTION AND PRESERVATION PROTOCOLS

SHREWS

<u>General</u>: Parasites, particularly the minuscule tapeworms characteristic of shrews, decompose rapidly.

Consequently, it is necessary to make special provisions for anticipated collections of insectivores. Live traps or pitfalls should be used, and should be checked at a minimum every 3 hours. This sampling schedule is necessary as tapeworms in shrews decompose starting about 2-3 hours following death of the host. Thus, traps must be checked frequently and any shrews collected need to be dissected immediately. Please note in field notes how fresh the shrew was at the time of dissection.

<u>Intestinal tracts</u>: materials must be as fresh as possible, as decomposition of parasite specimens in shrews is exceptionally rapid and the tapeworms are very tiny and delicate. Dissect the stomach and entire small intestine from the body, leave it intact. If the specimen is fresh and/or from a mature shrew, place it in a 1.8 mL nunc tube, or appropriate size (white caplet) and freeze in LN2. If the specimen isn't fresh or as space runs out the entire intact intestine can be preserved in ⁷⁰⁰/o ethanol, use a 20m1 scintillation vial. If ethanol is used it must be changed on the following day. <u>Immediate processing and preservation of intestinal tracts of shrews has the highest priority.</u>

OTHER MAMMALS

<u>General</u>: In water, using a petri dish of appropriate size, first uncoil the intestine, separate the small intestine, large intestine, and caecum, process separately. Record the total number (and sex ratio if possible) of each parasite type from each organ separately in the comments section of the AF sheets.

Small intestine: In water, remove the mesenteries, straighten, and open lengthwise starting in the anterior by carefully sliding blunt tipped iris scissors to cut open and expose the lumen. Alternatively, dissecting can be approached from the posterior end, allowing the dissector to encounter the posterior end of the worm first. Be careful not to cut tapeworms; remove intact tapes to a separate dish in water to relax. "Wash" (agitate) the intestine sections in the petri dish for the detached scolices and small nematodes. Record the location of the worm in the small intestine (first, second, or third part). Look carefully for any scolices if detached: this will be accomplished by using a dissecting scope or magnifying loop; pour extra water carefully away before this but do not lose the small worms.

<u>Caecum</u>: Nematodes or small flukes may be present in the caecum. Often trematodes (reddishbrownish in color) in the caecum of *Microtus* are covered with "mud" and are difficult to see and some nematodes are relatively small and obscure. It may be useful, therefore, to run material through a small sieve to first discard some fine particulates. *Dicrostonyx* has a nematode that is spiraled around the villi in the caecum wall.

Large Intestine: Process only if sufficient time is available; there will be little of importance here

in arvicolines. Use the same techniques as with the caecum.

ALL MAMMALS

<u>Stomach</u>: Open in a dish of water, examine for nematodes. These are usually associated with the lining of the stomach and are on the outside of the stomach contents.

<u>Lungs</u>: First remove the left half of the lungs for hanta virus (see later) and freeze: be careful and use sterilized forceps only (70% ethanol and cigarette lighter). Label nunc "Hanta". Visually examine rest of lungs for nematodes. These may appear as small tan lesions on the surface and extending deeper into the lung tissue. If nematodes are present freeze ½ lung in nunc tube (white caplet) and label Lung/Nematode. One rare genus, *Angiostrongylus*, can be found in the big arteries of lungs.

Bladder- Open bladder in petri dish and examine contents under dissecting microscope.

Other Organs/Tissues- Basically parasites can occur in almost any organ- generally they are most abundant in the GI tract, but other organs including the liver (and gall bladder), etc., and the body cavity, should be examined (see Gardner protocols). Liver cysts or other Taeiid larvae loose in the coelom or thoracic cavity should be preserved in 70% ethanol.

Important- remember that all dishes, and dissectin~ instruments have to be completely clean and dry between animals. Wash and then rinse with ethanol. Tips of probes, scissors and micro-forceps can also be passed briefly through a flame after dipping in ethanol.

PARASITE HANDLING AND PRESERVATION

<u>Cestodes</u> (All mammalian taxa excluding shrews): Following collection from the small intestine, each specimen should be held in filtered water for an extended period (minimum 2 hours, preferably more). This allows the tapeworm to fully relax, which is necessary to examine the internal structure of the proglottids. <u>Following relaxation and death in water, all strobilate adult tapeworms will be preserved in 70% ethanol</u>. Preservation should be done flat for large tapes including *Andrya* and some *Hymenolepis* in rodents; this is done by leaving the cestodes in a dish of ethanol overnight, and transferring the specimen to a vial the following day. Use the appropriate size vial for the specimen so there is sufficient preservative (a ratio of about 5:1 in volume for preservative relative to the specimen is maintained). The preservative should be changed once after 24 hours. Some tapeworms in *Microtus* are quite large (up to 20 cm), so be certain to use the proper size vial- one that is large enough for the worm and a sufficient amount of ethanol. Note the location of cestode in the intestine and record in the AF book. If problems with vial size, a big tape can cut in two parts and preserved in two vials. Use one number with a and b, mark on the notes.

<u>Digenia</u> (Flukes): Flukes can be relaxed in filtered water, which often allows specimens to expel eggs that might otherwise obscure some organs. Preserve flukes in 70% ethanol; (or alternatively freeze in LN2 (white caplet)); if there are large numbers do both. Keep parasites from different organ systems separate.

<u>Nematodes</u>: Nematodes should not be held in water for extended periods of time, as osmotic pressure will eventually cause the specimen to burst. Specimens should be washed in water or saline and then preserved in 70% ethanol or frozen in LN2 (white caplet); if there are large numbers do both. Keep parasites from different organ systems separate.

Enteric Coccidians (see protocols in Gardner): Fecal samples to isolate coccidia should be taken from all species of mammals. Collect a few pellets from the rectum or a scraping from the caecum/large intestine, crush the pellets and put in potassium dichromate (2% solution). **Important:** 1) do not overfill the vial, oxygen is necessary for survival of the coccida, and 2) the specimens should not be frozen.

Blood parasites

Spleen Smears: Divide the spleen in half. Prepare a spleen smear (see Gardner); air dry and then fix with 100% methanol; store in a dry container, avoid changes in temperatures, moisture and condensation. The focus for this work is *Clethrionomys, Microtus* and *Peromyscus* and shrews; any lagomorphs; and marmots. Do a smear from snap- trapped animals. Freeze 1/2 separately in Alsever's solution and include the other half with the heart. <u>Indicate on AF page if the spleen is enlarged</u>.

Brain tissue, *Marmota*: Collect some brain tissue for freezing (lavender caplet).

<u>Protocols for Ectoparasites</u> (see protocols in Gardner): Open collection bag and place in sealed jar with chloroformed cotton balls for 5 minutes or more. Loosely stroke pelage of animal into the bag, then examine more closely for ticks, fleas, and mites. Wash collection bag with 70% ethanol, then cut corner of bag and let contents drain into a small vial. Add ecto juice to fill. Comment in the AF book the condition of the animal (as to whether or not the animal was wet vs. dry). **Do not re-use collection bags!!**

<u>Protocols for Hanta Virus</u>: Focus on the following rodents: *Clethrionomys, Lemmus, Dicrostonyx* and *Microtus*. Be sterile. The rodent's left lobes of lungs are frozen in a single tube; do not include with other organs; mark tube as Hanta, use no caplet (these will go with Dr. Henttonen for later screening).

Tissue-Cyst Forming Coccidia: Typically these will be found in old arvicolines. *Sarcocystis* may be present on the peritoneum and in the musculature of the hind legs as whitish thread-like structures; if observed in the peritoneum, preserve some hind-leg musculature in 70% ethanol. *Frenkelia* may be present in the brain; cysts are easily seen as whitish spots (0.5-1.0 mm) on the surface of the brain. Do not collect from specimens with intact skulls destined for the Museum. In animals with broken skulls: first remove the upper part of the skull by cutting the bone between the eyes; remove skin from the eyes backwards; cut the skull (but not the brain) starting from the eyes along the sides, and then lift the top from the anterior part exposing the brain. If present, cysts (whitish spots) will be visible; remove brain with forceps; slice into 2 or 3 parts; preserve in 70% ethanol. Remember to save the dentition from these animals.

HOW TO FILL AND LABEL CRYOTUBES

In the Alaska Frozen Tissue Collection, tissue samples are stored in 1.8 milliliter plastic cryotubes. These should be **labeled with an ultrafine Nalgene lab marker no. 6310-0010** or a **ultra fine "Sharpie" permanent marker prior to cooling.** If the tubes are not labeled before cooling, it will be necessary to rewarm the tubes in order to write on them.

The standard tissues saved on birds and mammals are heart, kidney, liver, and spleen. For small species entire organs are often stored in one tube. For larger animals, only a subsample of the organ will fit in a tube. In some cases, muscle, skin, or blood may be the only tissues collected.

Specimens should be kept clean, but are not expected to be sterile. It is especially necessary to avoid cross contamination between individuals. Tiny amounts of DNA from another specimen can be amplified and corrupt results. Therefore, instruments and work surfaces should be cleaned after each individual is sampled. We use a ten percent solution of chlorine bleach in water to clean oft instruments. The instruments are wiped dry, then rinsed in clean water, and then wiped until dry with clean tissue paper. Bleach destroys DNA and is an excellent disinfectant. Alcohol preserves DNA and therefore should not be used to clean instruments.

COMMON PROBLEMS

Over filling: Tubes that contain too much tissue will split when the tissue freezes and expands. Observe the fill line (approximately 2/3 full) when preparing large samples.

<u>Loose caps</u>: Caps may come loose and the samples may come out of the tubes. Please tighten caps firmly. This is particularly important when tubes are traveling in Dewar flasks of liquid nitrogen.

<u>Inadequate labeling</u>: Sloppy handwriting and faulty writing implements are major problems. Write the AF number on the tube at least twice, and on the cap once. Don't try to write on greasy, wet, or frozen tubes. Writing may be worn off of tubes if they are subjected to a lot of agitation while traveling in liquid nitrogen. This problem occurs with bags used to presample the tissues as well. Bags should be clearly labeled and if possible, a label should be included in the bag with the sample. Be sure to avoid cross contamination among bagged tissues and try to collect samples large enough so that we can obtain a cleanly trimmed final sample.

ALASKA FROZEN TISSUE COLLECTION University of Alaska Museum

Preparator: Species: Clethring mys rutilus Sex: MER Country/State Alaska Quad: Charley River District (e.g. island, county, Nat'l Park): Yuch Specific locality: 12.5 Km ENE Kathul Mt. Latitude: 45°22.231 'N Longitude: 142°00.748 WAuthority: GPS Date of death; 24 July 2001 preparation: Nature of voucher (Circle one or more): skin skull skeleton fluid-preserved whole frozen tissues only other Preserved tissue #tubes pres Preserved tissue #tubes pres heart blood kidney karyotype heart & kidney ectoparasites 4000 H. Mag. spleen Panematode 1000 Liver & kidney Panematode 1	Collector: S.o					_
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